

CLAIMS

The following is a complete listing of the claims.

1. (Original) A method of comparing protein compositions between at least two different samples comprising:

(a) preparing an extract of proteins from each of said at least two samples;
(b) providing a set of matched luminescent dyes chosen from dyes capable of covalently binding to proteins within said extract of proteins, wherein each dye within said set

(1) has a net charge which will maintain the overall net charge of the proteins upon such covalent binding and has ionic and pH characteristics whereby relative migration of a protein labeled with any one of said dyes is the same as relative migration of said protein labeled with another dye in said set,

(2) emits luminescent light at a wavelength that is sufficiently different from the emitted luminescent light of remaining dyes in said set to provide a detectably different light signal;

(c) reacting each extract of proteins of step (a) with a different dye from said set of step (b) to provide dye-labeled proteins;

(d) mixing each of said dye labeled proteins to form a single mixture of different dye-labeled proteins;

(e) separating the dye-labeled proteins of interest within said mixture; and

(f) detecting the difference in luminescent intensity between the different dye-labeled proteins of interest by:

capturing images of the dye-labeled proteins at different wavelengths of emitted luminescence; and

processing the images to determine the difference in luminescent intensity.

2. (Original) The method of claim 1, wherein said samples are cell samples.

3. (Original) The method of claim 1, wherein separating the dye-labeled proteins is by an electrophoretic method.

4. (Original) The method of claim 1, wherein said capturing and processing steps are performed on at least a first and a second image.

5. (Original) The method of claim 1, wherein processing the images includes processing the images with a computer.

6. (Original) The method of claim 1, wherein processing the images includes performing arithmetic operations on values representative of pixel intensities in the images.

7. (Original) The method of claim 1, wherein capturing the images includes: capturing a first image using a first filter or filters that only allows the passage of light having the wavelength of the luminescent light emitted by a first dye used to label the proteins; and

capturing a second image using a second filter or filters that only allows the passage of light having the wavelength of the luminescent light emitted by a second dye used to label the proteins.

8. (Original) The method of claim 7, wherein processing the first and second images includes subtracting the first image from the second image.

9. (Original) The method of claim 8, wherein processing the first and second images further includes multiplying one of the first and second image by a fluorescence balancing factor prior to subtracting the first image from the second image.

10. (Original) The method of claim 7, wherein processing the first and second images includes dividing the first image by the second image.

11. (Original) The method of claim 10, wherein processing the first and second images further includes normalizing the first and second images to a common intensity range prior to dividing the first image by the second image.

12. (Original) The method of claim 11, wherein processing the first and second images further includes multiplying one of the first and second images by a fluorescence balancing factor.

13. (Original) A method of comparing protein compositions between at least two different samples comprising:

(a) preparing an extract of proteins from each of said at least two samples;
(b) providing a set of matched luminescent dyes chosen from dyes capable of covalently binding to proteins within said extract of proteins, wherein each dye within said set

(1) has a net charge which will maintain the overall net charge of the proteins upon such covalent binding and has ionic and pH characteristics whereby relative migration of a protein labeled with any one of said dyes is the same as relative migration of said protein labeled with another dye in said set,

(2) emits luminescent light at a wavelength that is sufficiently different from the emitted luminescent light of remaining dyes in said set to provide a detectably different light signal;

(c) reacting each extract of proteins of step (a) with a different dye from said set of step (b) to provide dye-labeled proteins;
(d) mixing each of said dye labeled proteins to form a single mixture of different dye-labeled proteins;
(e) separating the dye-labeled proteins of interest within said mixture; and
(f) detecting the difference in luminescent intensity between the different dye-labeled proteins of interest by:

capturing a first image of the dye labeled proteins using a first filter or filters that only allows the passage of light having the wavelength of the luminescent light emitted by a first dye used in labeling the proteins of interest;

capturing a second image of the dye labeled proteins using a second filter or filters that only allows the passage of light having the wavelength of the luminescent light emitted by a second dye used in labeling the proteins of interest; and
processing the first and second images to determine the difference in luminescent intensity.

14. (Original) The method of claim 13, wherein said samples are cell samples.

15. (Original) The method of claim 14, wherein processing the first and second images includes processing the first and second images with a computer.

16. (Original) The method of claim 13, wherein processing the first and second images includes performing arithmetic operations on values representative of pixel intensities in the first and second images.

17. (Original) The method of claim 16, wherein processing the first and second images includes subtracting the first image from the second image.

18. (Original) The method of claim 17, wherein processing the first and second images further includes multiplying one of the first and second image by a fluorescence balancing factor prior to subtracting the first image from the second image.

19. (Original) The method of claim 16, wherein processing the first and second images includes dividing the first image by the second image.

20. (Original) The method of claim 19, wherein processing the first and second images further includes normalizing the first and second images to a common intensity range prior to dividing the first image by the second image.

21. (Original) The method of claim 20, wherein processing the first and second images further includes multiplying one of the first and second images by a fluorescence balancing factor.

22. (Original) The method of claim 13, wherein separating the dye-labeled proteins is by an electrophoretic method.

23. (Withdrawn) A method of comparing protein compositions between at least two different samples comprising:

(a) preparing an extract of proteins from each of said at least two samples;
(b) providing a set of matched luminescent dyes chosen from dyes capable of covalently binding to proteins within said extract of proteins, wherein each dye within said set

(1) has a net charge which will maintain the overall net charge of the proteins upon such covalent binding and has ionic and pH characteristics whereby relative migration of a protein labeled with any one of said dyes is the same as relative migration of said protein labeled with another dye in said set,

(2) emits luminescent light at a wavelength that is sufficiently different from the emitted luminescent light of remaining dyes in said set to provide a detectably different light signal;

(c) reacting each extract of proteins of step (a) with a different dye from said set of step (b) to provide dye-labeled proteins;

(d) mixing each of said dye labeled proteins to form a single mixture of different dye-labeled proteins;

(e) placing said mixture in an electrophoresis gel and separating the dye-labeled proteins of interest within said mixture;

(f) making images of the gel; and,

(g) processing the images with a computer to detect the difference in luminescent intensity between the different dye-labeled proteins of interest.

24. (Withdrawn) The method of claim 23, wherein said samples are cell samples.

25. (Withdrawn) A method of comparing protein compositions between at least two different samples comprising:

(a) preparing an extract of proteins from each of said at least two samples;
(b) providing a set of matched luminescent dyes chosen from dyes capable of covalently binding to proteins within said extract of proteins, wherein each dye within said set

(1) has a net charge which will maintain the overall net charge of the proteins upon such covalent binding and has ionic and pH characteristics whereby relative migration of a protein labeled with any one of said dyes is the same as relative migration of said protein labeled with another dye in said set,

(2) emits luminescent light at a wavelength that is sufficiently different from the emitted luminescent light of remaining dyes in said set to provide a detectably different light signal;

(c) reacting each extract of proteins of step (a) with a different dye from said set of step

(b) to provide dye-labeled proteins;

(d) mixing each of said dye labeled proteins to form a single mixture of different dye-labeled proteins;

(e) separating the dye-labeled proteins of interest within said mixture;

(f) capturing images of said separated dye-labeled proteins; and,

(g) detecting the difference in luminescent intensity between the different dye-labeled proteins of interest by computer analysis of the images.

26. (Withdrawn) The method of claim 25, wherein said samples are cell samples.

27. (Withdrawn) The method of claim 25, wherein detecting the difference in luminescent intensity further comprises:

capturing first and second images of the dye-labeled proteins; and

performing arithmetic operations on values representative of pixel intensities in the first and second images.

28. (Withdrawn) The method of claim 27, further comprising subtracting the first image from the second image.

29. (Withdrawn) The method of claim 28, further comprising multiplying one of the first and second image by a fluorescence balancing factor prior to subtracting the first image from the second image.

30. (Withdrawn) The method of claim 29, further comprising dividing the first image by the second image.

31. (Withdrawn) The method of claim 30, further comprising normalizing the first and second images to a common intensity range prior to dividing the first image by the second image.

32. (Withdrawn) The method of claim 31, further comprising multiplying one of the first and second images by a fluorescence balancing factor.

33. (Withdrawn) A method of comparing protein compositions between at least two different samples comprising:

- (a) preparing an extract of proteins from each of said at least two samples;
- (b) providing a set of matched luminescent dyes chosen from dyes capable of covalently binding to proteins within said extract of proteins, wherein each dye within said set

- (1) has a net charge which will maintain the overall net charge of the proteins upon such covalent binding and has ionic and pH characteristics whereby relative migration of a protein labeled with any one of said dyes is the same as relative migration of said protein labeled with another dye in said set,

(2) emits luminescent light at a wavelength that is sufficiently different from the emitted luminescent light of remaining dyes in said set to provide a detectably different light signal;

(c) reacting each extract of proteins of step (a) with a different dye from said set of step (b) to provide dye-labeled proteins;

(d) mixing each of said dye labeled proteins to form a single mixture of different dye-labeled proteins;

(e) separating the dye-labeled proteins of interest within said mixture; and

(f) detecting the difference in luminescent intensity between the different dye-labeled proteins of interest by:

capturing luminescent data for the dye-labeled proteins at different wavelengths of emitted luminescence; and

processing said data to determine the difference in luminescent intensity.

34. (Withdrawn) The method of claim 33, wherein said samples are cell samples.

35. (Withdrawn) The method of claim 33, wherein the dye-labeled proteins are separated by chromatography.

36. (Withdrawn) The method of claim 35, wherein capturing luminescent data comprises passing dye-labeled protein through a fluorimeter and measuring the relative fluorescent intensity of the dye-labeled proteins.

37. (Withdrawn) The method of claim 33, wherein processing the data includes processing the data with a computer.

38. (New) A method of comparing protein compositions between at least two different samples comprising:

(a) preparing an extract of proteins from each of said at least two samples;

(b) providing a set of matched luminescent dyes selected from the group consisting of

cyanine dyes, dipyrromethene boron difluoride dyes, squarate dyes and derivatives thereof, said dyes being capable of covalently binding to proteins within said extract of proteins, wherein each dye within said set

(1) has a net charge which will maintain the overall net charge of the proteins upon such covalent binding and has ionic and pH characteristics whereby relative migration of a protein labeled with any one of said dyes is the same as relative migration of said protein labeled with another dye in said set,

(2) emits luminescent light at a wavelength that is sufficiently different from the emitted luminescent light of remaining dyes in said set to provide a detectably different light signal;

(c) reacting each extract of proteins of step (a) with a different dye from said set of step (b) to provide dye-labeled proteins;

(d) mixing each of said dye labeled proteins to form a single mixture of different dye-labeled proteins;

(e) separating the dye-labeled proteins of interest within said mixture; and

(f) detecting the difference in luminescent intensity between the different dye-labeled proteins of interest by:

capturing images of the dye-labeled proteins at different wavelengths of emitted luminescence; and

processing the images to determine the difference in luminescent intensity.

39. (New) A method of comparing protein compositions between at least two different samples comprising:

(a) preparing an extract of proteins from each of said at least two samples;

(b) providing a set of matched luminescent dyes selected from the group consisting of cyanine dyes, dipyrromethene boron difluoride dyes, squarate dyes and derivatives thereof, said dyes being capable of covalently binding to proteins within said extract of proteins, wherein each dye within said set

(1) has a net charge which will maintain the overall net charge of the proteins upon such covalent binding and has ionic and pH characteristics whereby relative migration of a protein labeled with any one of said dyes is the same as relative

migration of said protein labeled with another dye in said set,

(2) emits luminescent light at a wavelength that is sufficiently different from the emitted luminescent light of remaining dyes in said set to provide a detectably different light signal;

(c) reacting each extract of proteins of step (a) with a different dye from said set of step

(b) to provide dye-labeled proteins;

(d) mixing each of said dye labeled proteins to form a single mixture of different dye-labeled proteins;

(e) separating the dye-labeled proteins of interest within said mixture; and

(f) detecting the difference in luminescent intensity between the different dye-labeled proteins of interest by:

capturing a first image of the dye labeled proteins using a first filter or filters that only allows the passage of light having the wavelength of the luminescent light emitted by a first dye used in labeling the proteins of interest;

capturing a second image of the dye labeled proteins using a second filter or filters that only allows the passage of light having the wavelength of the luminescent light emitted by a second dye used in labeling the proteins of interest; and

processing the first and second images to determine the difference in luminescent intensity.